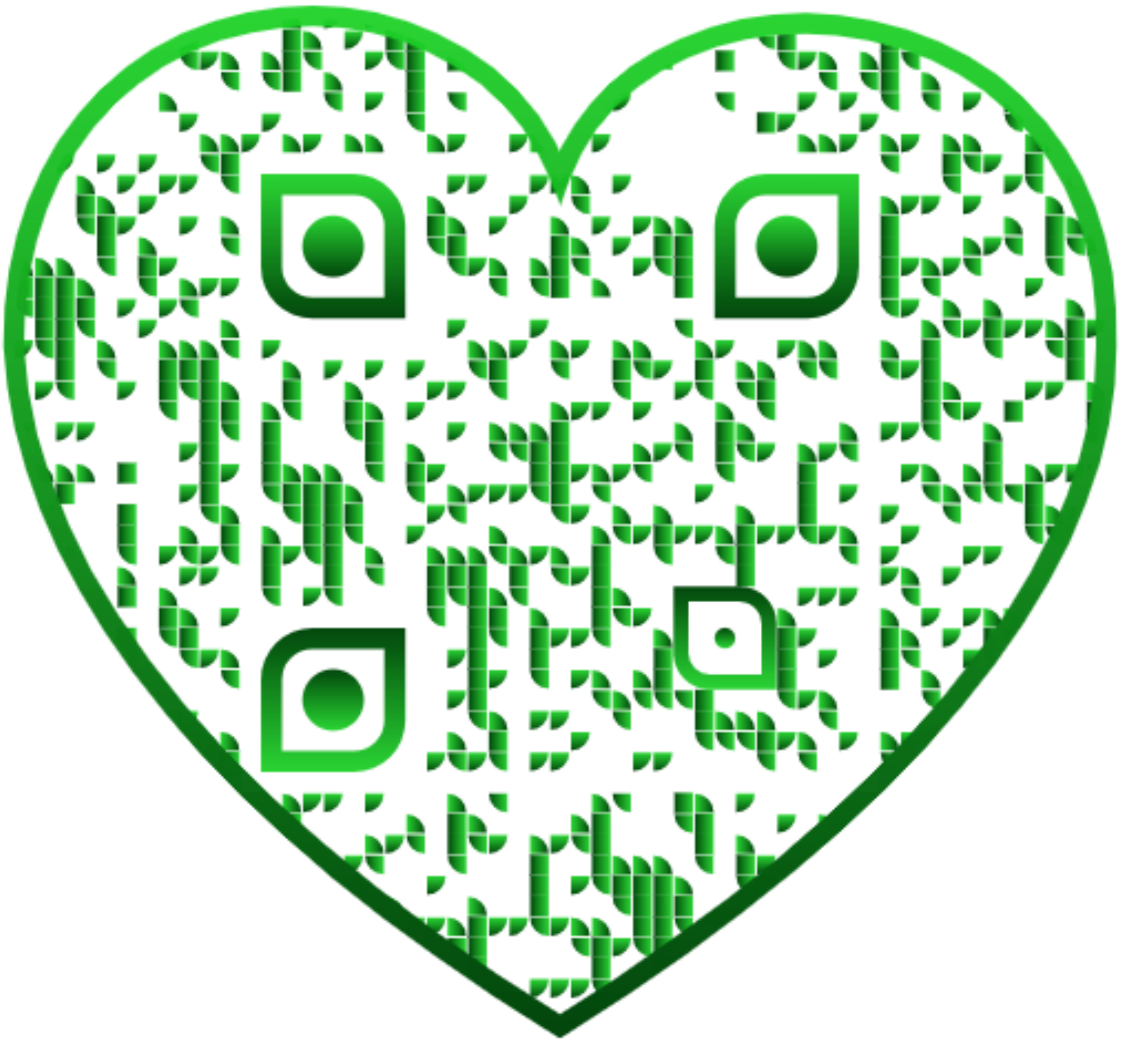
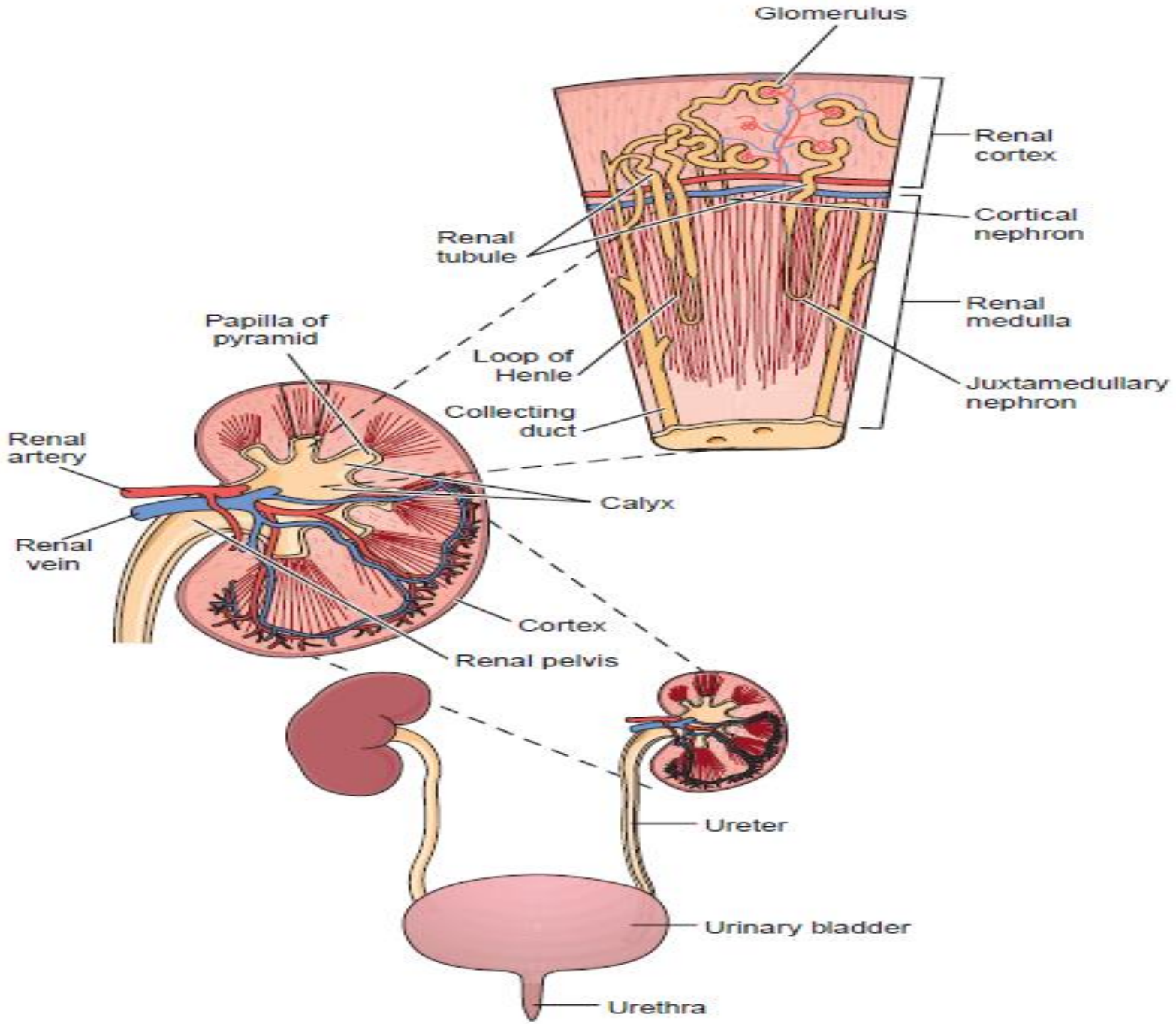
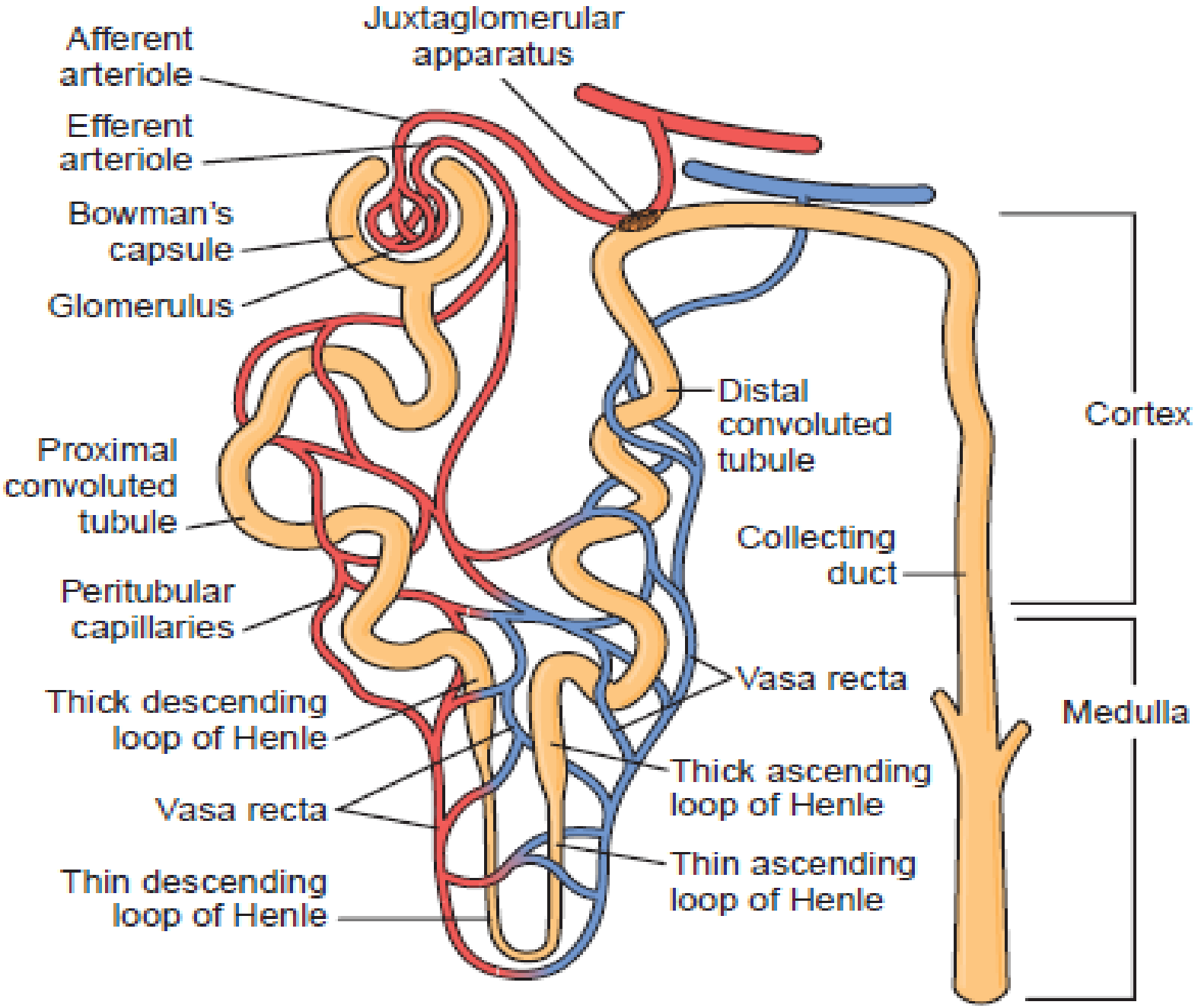
Urinalysis

- Urine is formed in the kidneys, is a product of ultrafiltration of plasma by the renal glomeruli.




Glomerulus
Renal cortex
Cortical nephron
Renal tubule
Renal medulla
Papilla of pyramid
Loop of Henle
Juxtamedullary nephron
Collecting duct
Renal artery
Calyx
Renal vein
Cortex
Renal pelvis
Ureter
Urinary bladder
Urethra


Afferent arteriole
Juxtaglomerular apparatus
Efferent arteriole
Bowman's capsule
Glomerulus
Distal convoluted tubule
Cortex
Proximal convoluted tubule
Collecting duct
Peritubular capillaries
Vasa recta
Medulla
Thick descending loop of Henle
Thick ascending loop of Henle
Vasa recta
Thin ascending loop of Henle
Thin descending loop of Henle

Importance of urine analysis

- It can detect diseases which pass unnoticed.

For example, D.M, chronic UTI.

- Diagnosis of many renal diseases.

As nephrotic, nephritic syndrome, acute renal failure,

multiple myeloma

Table 3–4 Types of Urine Specimens

Type of Specimen	Purpose
Random	Routine screening
First morning	Routine screening Pregnancy tests Orthostatic protein
Fasting (second morning)	Diabetic screening/monitoring
2-hour postprandial	Diabetic monitoring
Glucose tolerance test	Optional with blood samples in glucose tolerance test
24-h (or timed)	Quantitative chemical tests
Catheterized	Bacterial culture
Midstream clean-catch	Routine screening Bacterial culture
Suprapubic aspiration	Bladder urine for bacterial culture Cytology
Three-glass collection	Prostatic infection

Specimen Collection

- First morning voiding (most concentrated)
- Record collection time
- Type of specimen (e.g. “clean catch”)
- Analyzed within 2 hours of collection
- Free of debris or vaginal secretions

Urine composition

- Urine, a very complex fluid, is composed of 95% water and 5% solids .It is the end product of the metabolism carried out by billions of cells and results in an average urinary out put of 1-1.5 L per day.
- Almost all substances found in urine are also find in the blood although in different concentration.
- Urine may also contain formed elements such as cells, casts, crystals, mucus and bacteria.

Table 3–1 Composition of Urine Collected for 24 Hours

Component	Amount	Remark
Organic		
Urea	25.0–35.0 g	60%–90% of nitrogenous material; derived from the metabolism of amino acids into ammonia
Creatinine	1.5 g	Derived from creatine, nitrogenous substance in muscle tissue
Uric acid	0.4–1.0 g	Common component of kidney stones; derived from catabolism of nucleic acid in food and cell destruction
Hippuric acid	0.7 g	Benzoic acid is eliminated from the body in this form; increases with high-vegetable diets
Other substances	2.9 g	Carbohydrates, pigments, fatty acids, mucin, enzymes, hormones; may be present in small amounts depending on diet and health
Inorganic		
Sodium chloride (NaCl)	15.0 g	Principal salt; varies with intake
Potassium (K^+)	3.3 g	Occurs as chloride, sulfate, and phosphate salts
Sulfate (SO_4^{2-})	2.5 g	Derived from amino acids
Phosphate (PO_4^{3-})	2.5 g	Occurs primarily as sodium compounds that serve as buffers in the blood
Ammonium (NH_4^+)	0.7 g	Derived from protein metabolism and glutamine in kidneys; amount varies depending on blood and tissue fluid acidity
Magnesium ($Mg2^+$)	0.1 g	Occurs as chloride, sulfate, phosphate salts
Calcium ($Ca2^+$)	0.3 g	Occurs as chloride, sulfate, phosphate salts

Types of Analysis

- Macroscopic Examination
- Chemical Analysis (Urine Dipstick)
- Microscopic Examination
- Culture (not covered in this lecture)
- Cytological Examination

Macroscopic Examination

Volume

Color

Odour

Appearance

Urinary volume

1-volume:

Normal = 600-1550ml

Polyuria- >2000ml

Oliguria-<400ml

Anuria-complete cessation of urine(<200ml)

Nocturia-excretion of urine by a adult of >500ml with a specific gravity of <1.018 at night (characteristic of chronic glomerulonephritis)

Causes of polyuria

Diabetes mellitus
Diabetes insipidus
Polycystic kidney
Chronic renal failure
Diuretics
Intravenous saline/glucose

oliguria

Dehydration-vomiting, diarrhoea, excessive sweating

Renal ischemia

Acute tubular necrosis

Obstruction to the urinary tract

Acute renal failure

2-Odor:

– Ammonia-like:	(Urea-splitting bacteria)
– Foul, offensive:	Old specimen, pus or inflammation
– Sweet:	Glucose
– Fruity:	Ketones
– Maple syrup-like:	Maple Syrup Urine Disease1-

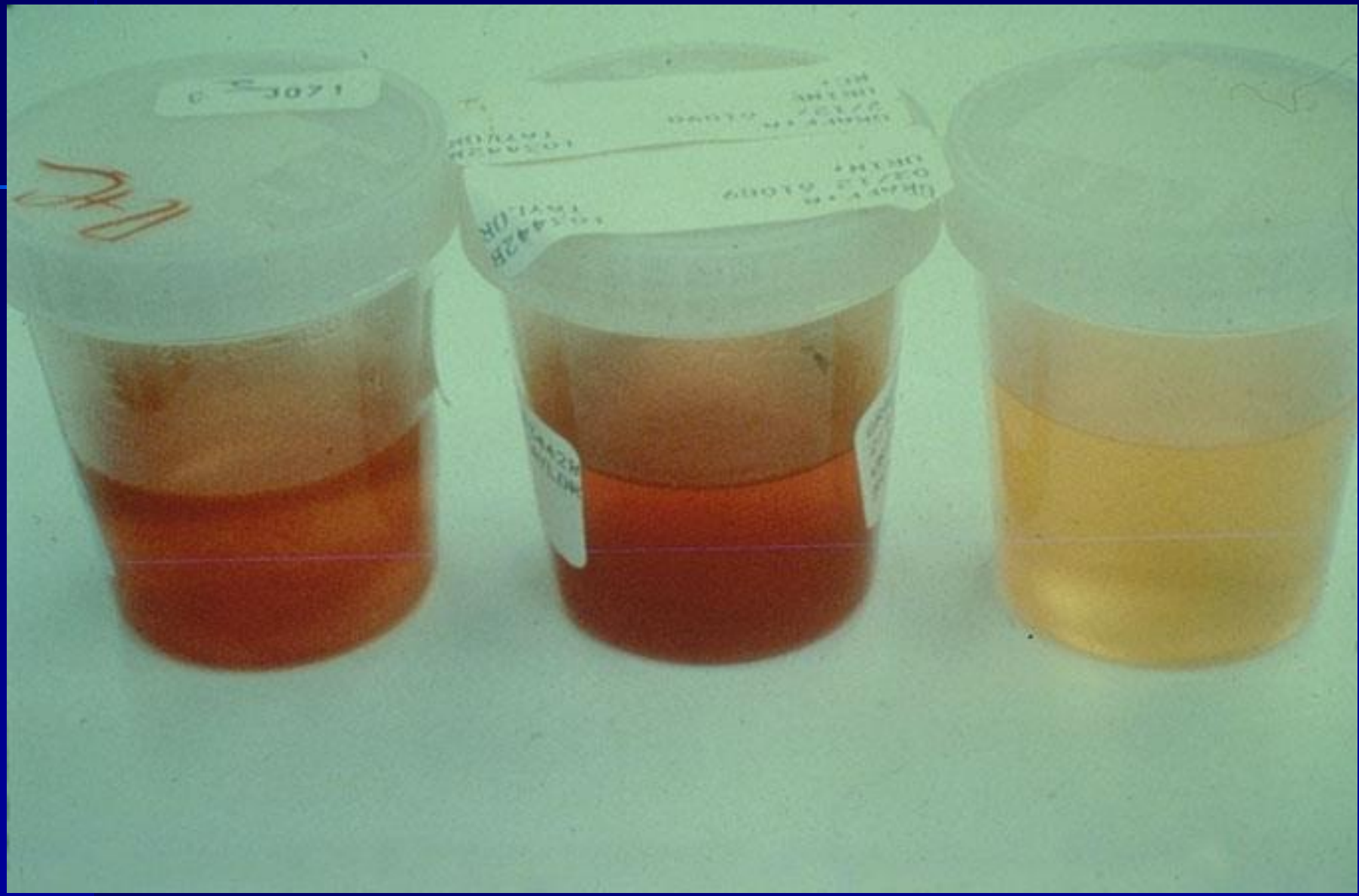
3-Color:

– Colorless	Diluted urine
– Deep Yellow	Concentrated Urine, Riboflavin
– Yellow-Green	Bilirubin / Biliverdin
– Red	Blood / Hemoglobin
– Brownish-red	Acidified Blood (Actute GN)
– Brownish-black	Homogentisic acid (Melanin)

Table 4–1 Laboratory Correlation of Urine Color[11]

Color	Cause	Clinical/Laboratory Correlations
Colorless	Recent fluid consumption	Commonly observed with random specimens
Pale yellow	Polyuria or diabetes insipidus	Increased 24-hour volume
	Diabetes mellitus	Elevated specific gravity and positive glucose test result
	Dilute random specimen	Recent fluid consumption
Dark yellow	Concentrated specimen	May be normal after strenuous exercise or in first morning specimen
Amber		Deydration from fever or burns
Orange	Bilirubin	Yellow foam when shaken and positive chemical test results for bilirubin
	Acriflavine	Negative bile test results and possible green fluorescence
	Phenazopyridine (Pyridium)	Drug commonly administered for urinary tract infections May have orange foam and thick orange pigment that can obscure or interfere with reagent strip readings
	Nitrofurantoin	Antibiotic administered for urinary tract infections
	Phenindione	Anticoagulant, orange in alkaline urine, colorless in acid urine
Yellow-green Yellow-brown	Bilirubin oxidized to biliverdin	Colored foam in acidic urine and false-negative chemical test results for bilirubin
Green	*Pseudomonas* infection	Positive urine culture
Blue-green	Amitriptyline	Antidepressant
	Methocarbamol (Robaxin)	Muscle relaxant, may be green-brown
	Clorets	None
	Indican	Bacterial infections
	Methylene blue	Fistulas
	Phenol	When oxidized

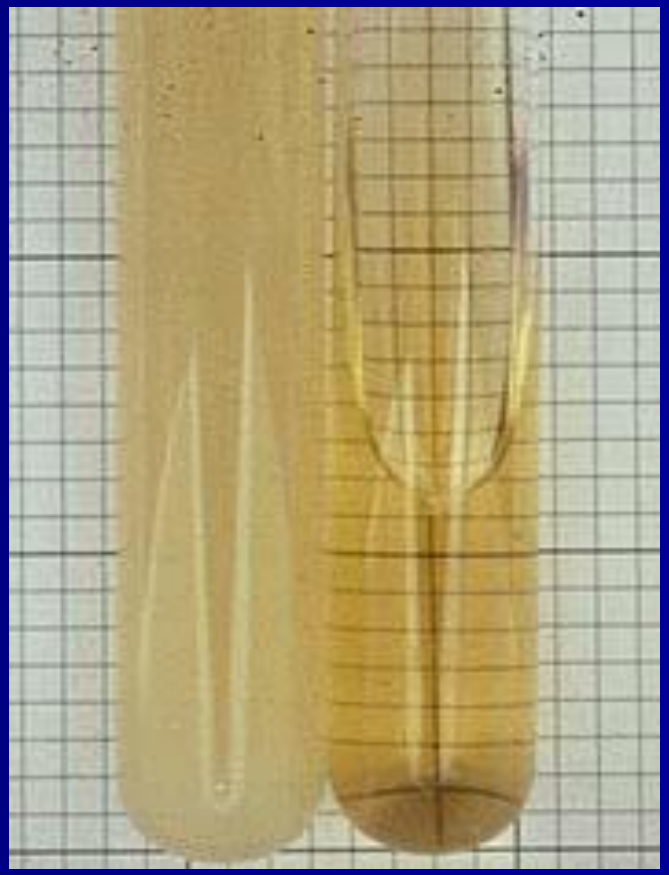
Color	Cause	Clinical/Laboratory Correlations
Pink Red	RBCs	Cloudy urine with positive chemical test results for blood and RBCs visible microscopically
	Hemoglobin	Clear urine with positive chemical test results for blood; intravascular hemolysis
	Myoglobin	Clear urine with positive chemical test results for blood; muscle damage
	Porphyrins	Negative chemical test results for blood Detect with Watson-Schwartz screening test or fluorescence under ultraviolet light
	Beets	Alkaline urine of genetically susceptible persons
	Rifampin	Tuberculosis medication
	Menstrual contamination	Cloudy specimen with RBCs, mucus, and clots
Brown Black	RBCs oxidized to methemoglobin	Seen in acidic urine after standing; positive chemical test result for blood
	Methemoglobin	Denatured hemoglobin
	Homogentisic acid (alkaptonuria)	Seen in alkaline urine after standing; specific tests are available
	Melanin or melanogen	Urine darkens on standing and reacts with nitroprusside and ferric chloride
	Phenol derivatives	Interfere with copper reduction tests
	Argyrol (antiseptic)	Color disappears with ferric chloride
	Methyldopa or levodopa	Antihypertensive
	Metronidazole (Flagyl)	Darkens on standing



Clarity (Transparency).

Normal urine clear or transparent, any turbidity will indicate.

- WBCs (pus).
- RBCs
- Epithelial cells
- Bacteria
- Casts
- Crystals
- Lymph
- Semen.



Appearance

Table 4–2 Urine Clarity

Clarity	Term
Clear	No visible particulates, transparent.
Hazy	Few particulates, print easily seen through urine.
Cloudy	Many particulates, print blurred through urine.
Turbid	Print cannot be seen through urine.
Milky	May precipitate or be clotted.

Chemical Analysis

Chemical Analysis

Dipstick reagent strip method

chemical-impregnated absorbent pads attached to a plastic strip.

Glucose

Bilirubin

Ketones

Specific Gravity

Blood

pH

Protein

Urobilinogen

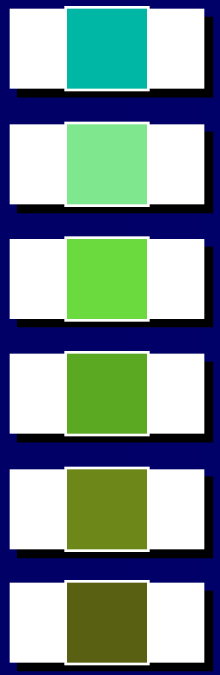
Nitrite

Leukocyte Esterase

A color-producing chemical reaction takes place when the absorbent pad comes in contact with urine.

The Urine Dipstick:

Glucose



Negative

Trace (100 mg/dL)

+ (250 mg/dL)

++ (500 mg/dL)

+++ (1000 mg/dL)

++++ (2000+ mg/dL)

Chemical Principle

Glucose Oxidase

$$\text{Glucose} + 2\ H_2O + O_2 \text{ ---> } \text{Gluconic Acid} + 2\ H_2O_2$$

Horseradish Peroxidase

$$3\ H_2O_2 + KI \text{ ---> } KIO_3 + 3\ H_2O$$

Read at 30 seconds
RR: Negative

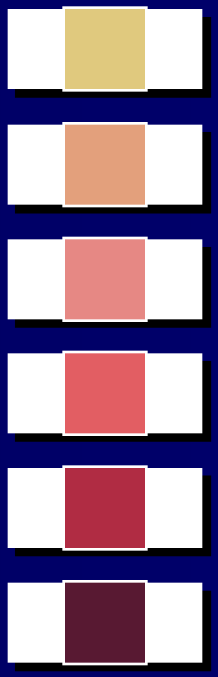
Uses of Urine Glucose Detection

Significance

- Diabetes mellitus.
- Renal glycosuria.
- Pancreatic cancer.
- Acromegaly.
- Cushing syndrome.
- Hyperthyrodism.
- Stress.
- Gestional diabetes

The Urine Dipstick:

Ketones



Negative

Trace (5 mg/dL)

+ (15 mg/dL)

++ (40 mg/dL)

+++ (80 mg/dL)

++++ (160+ mg/dL)

Chemical Principle

Acetoacetic Acid + Nitroprusside
------> Colored Complex

Read at 40 seconds
RR: Negative

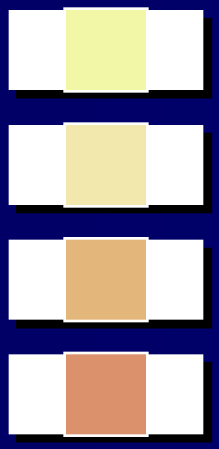
Uses and Limitations of Urine Ketone Detection

Significance

- Diabetic ketoacidosis
- Prolonged fasting
  Starvation
  Vomiting
  Malabsorption

The Urine Dipstick:

Bilirrubin



Negative

\+ (weak)

++ (moderate)

+++ (strong)

Chemical Principle

Bilirubin + Diazo salt ---*Acidic*---> Azobilirubin

Read at 30 seconds
RR: Negative

Uses of Urine Bilirubin Detection

-

Hydrophobic substance difficult to excretion and easy reabsorption.

Its appearance in urine abnoramal.

Significance

Hepatitis.

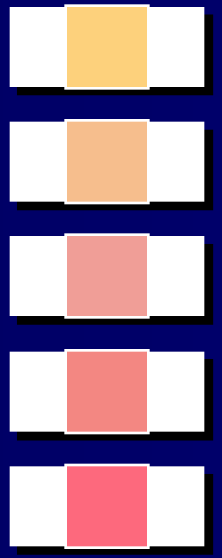
Cirrhosis

Liver disorder.

Biliary obstruction.

The Urine Dipstick:

Urobilinogen



0.1 mg/dL

1 mg/dL

2 mg/dL

4 mg/dL

8 mg/dL

Chemical Principle

Urobilinogen + Diethylaminobenzaldehyde
(Ehrlich's Reagent)

-------> Colored Complex

Read at 60 seconds

RR: 0.02-1.0 mg/dL

Uses of Urobilinogen Detection

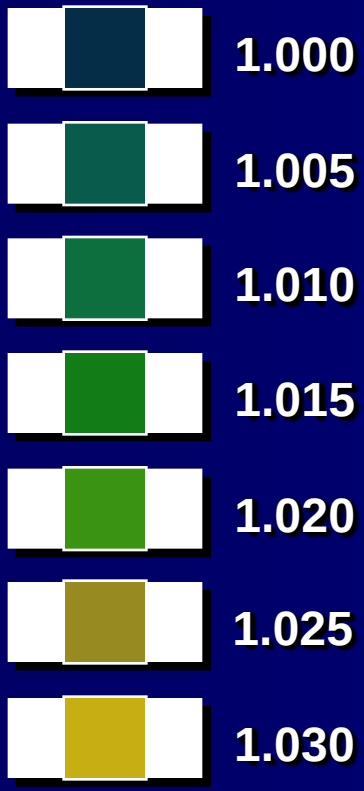
Significance

- High: increased hepatic processing of bilirubin
- Low: bile obstruction

The Urine Dipstick:

Specific Gravity

-Specific gravity is defined as the *density* of a solution compared with the density of a similar volume of distilled water at a similar temperature.
-ability of the kidneys to selectively reabsorb essential chemicals and water from the glomerular filtrate



Chemical Principle

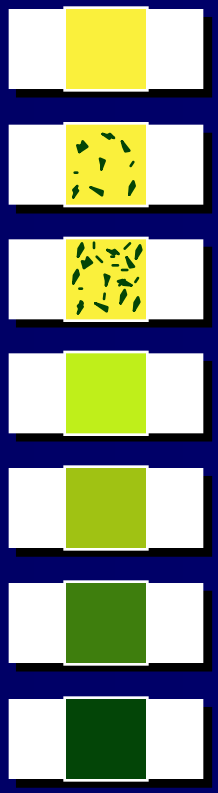
X^+ + Polymethyl vinyl ether / maleic anhydride
---------->
X^+-Polymethyl vinyl ether / maleic anhydride + H^+

H^+ interacts with a Bromthymol Blue indicator to form a colored complex.

Read up to 2 minutes
RR: 1.003-1.035

The Urine Dipstick:

Blood



Negative

Trace (non-hemolyzed)

Moderate (non-hemolyzed)

Trace (hemolyzed)

+ (weak)

++ (moderate)

+++ (strong)

Chemical Principle

Lysing agent to lyse red blood cells

Diisopropylbenzene dihydroperoxide + Tetramethylbenzidine

Heme ----------> Colored Complex

Read at 60 seconds
RR: Negative
Analytic Sensitivity: 10 RBCs

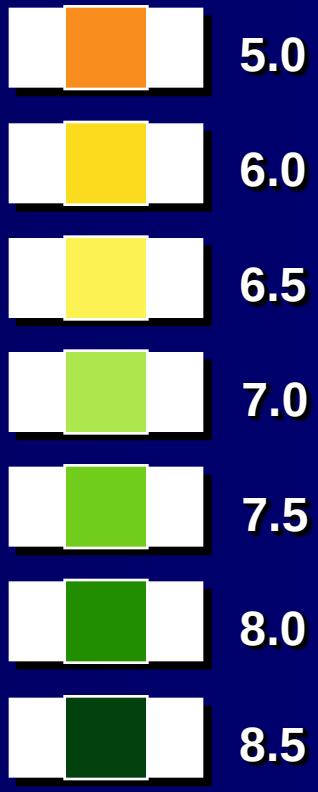
Uses of Urine Blood Detection

Significance

- Hematuria (nephritis, trauma, etc)
- Hemoglobinuria (hemolysis, etc)
- Myoglobinuria (rhabdomyolysis, etc)

The Urine Dipstick:

pH



Chemical Principle

H^+ interacts with:
Methyl Red (at high concentration; low pH) and Bromthymol Blue (at low concentration; high pH), to form a colored complexes (dual indicator system)

Read up to 2 minutes
R.R.: 4.5-8.0

Uses and Limitations of Urine pH Detection

Significance

- Acidic (less than 6.7): metabolic acidosis, high-protein diet , Diabetes mellitus, Dehydration, Medication.

- Alkaline (greater than 8.0): renal tubular acidosis, Hyperventilation, Vomiting , presence of urese producing bacteria.

The Urine Dipstick:

Protein

Negative

Trace

+ (30 mg/dL)

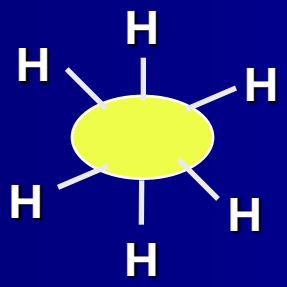
++ (100 mg/dL)

+++ (300 mg/dL)

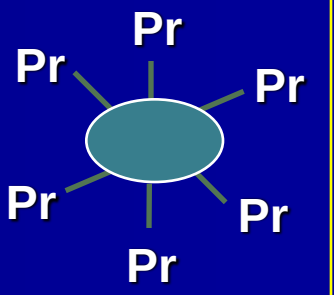
++++ (2000 mg/dL)

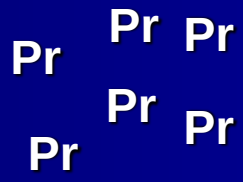
Chemical Principle

"Protein Error of Indicators Method"



Tetrabromphenol Blue
(buffered to pH 3.0)





Read at 60 seconds
RR: Negative

Proteins in "Normal" Urine

Protein	% of Total	Daily Maximum
Albumin	40%	60 mg
Tamm-Horsfall	40%	60 mg
Immunoglobulins	12%	24 mg
Secretory IgA	3%	6 mg
Other	5%	10 mg
TOTAL	100%	150 mg

Causes of Proteinuria

Renal

- Glomerulonephritis
- Nephrotic syndrome
- Renal tumor or infection

Pre-Renal

- Fever
- Renal hypoxia
- Hypertension

Post-Renal

- Cystitis
- Urethritis or prostatitis
- Contamination with vaginal secretions

Uses of Urine Protein Detection

Significance

nephrotic syndrome.

The Urine Dipstick:

Nitrite

Negative

Positive

Chemical Principle

Nitrite + p-arsenilic acid --(*Acidic*)--> Diazo compound

Diazo compound + Tetrahydrobenzoquinolinol ----------> Colored Complex

Read at 60 seconds
RR: Negative

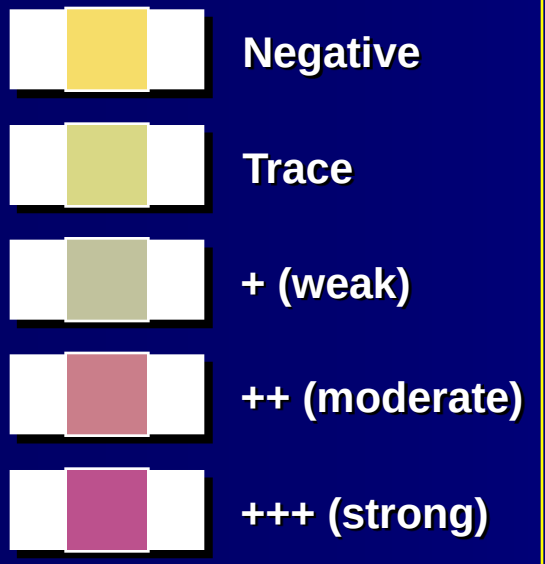
Uses of Nitrite Detection

Significance

- Gram negative bacteriuria

The Urine Dipstick:

Leukocyte Esterase



Chemical Principle

Derivatized pyrrole amino acid ester

Esterases ----------> 3-hydroxy-5-phenyl pyrrole

3-hydroxy-5-phenyl pyrrole + diazo salt ----------> Colored Complex

Read at 2 minutes
RR: Negative
Analytic Sensitivity: 3-5 WBCs

Uses Leukocyte Esterase Detection

Significance

- Pyuria
- Acute inflammation
- Renal calculus